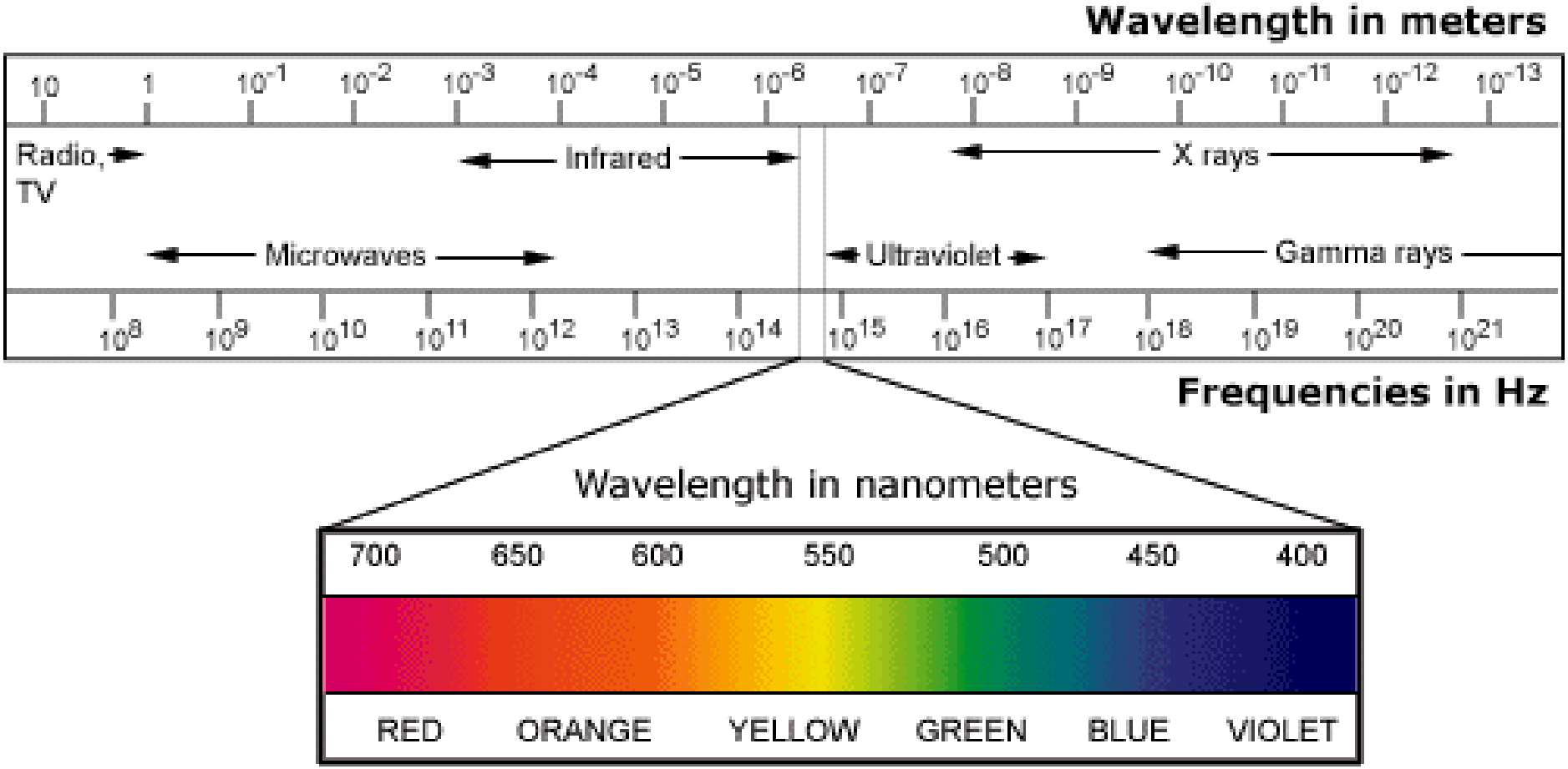
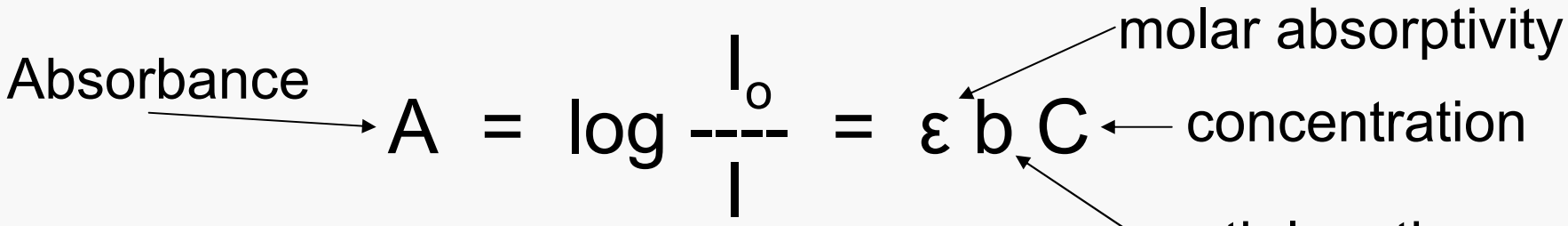
Spin States	Molecular Rotations	Molecular Vibrations	Outer Shell Electrons	Inner Shell Electrons	Nuclear Transitions
NMR EPR	Microwave Absorption Spectroscopy	Infrared Absorption Spectroscopy	UV-vis Absorption, Fluorescence	X-Ray Absorption, Fluorescence	Gamma Ray Spectroscopy



R O Y G B V

Quantitative Aspects of Absorption

Beer-Lambert Law (or Beer's Law)



Transmittance

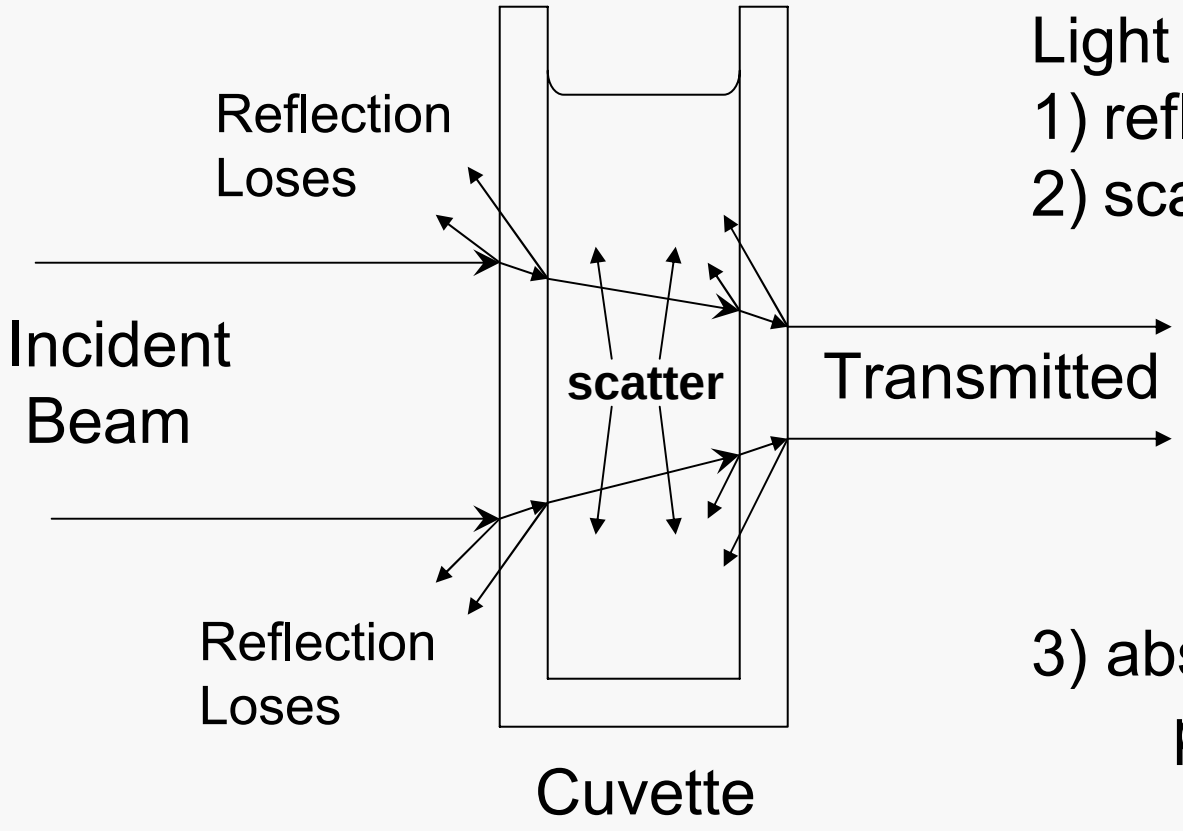
$$T = \frac{I}{I_o} \qquad \%T = T \times 100$$

I_o = measured source intensity

I = measured intensity after absorption

Intensity change does not change absorbance

Effects other than absorption that reduce source intensity (i.e., scattering, reflection) may also be measured as absorbance and must be accounted for when measuring I & I_o



Light loses occur due to:
1) reflection at boundaries
2) scattering by molecules or particles
3) absorption which is process of interest

- Absorbance & Transmittance are unitless
- If C is mol/L & b is in cm then ε is L/mol-cm
- To minimize the effect of light loses from reflection the procedure followed in UV-vis spectrophotometry is to measure I_o with a reference blank of pure solvent in the light path & then measure I under the same conditions – cuvettes should be optically matched if using 2 & clean, free of scratches, lint, fingerprints, etc.

Beer's Law applies to all absorption processes

Assumptions made in deriving Beer's Law:

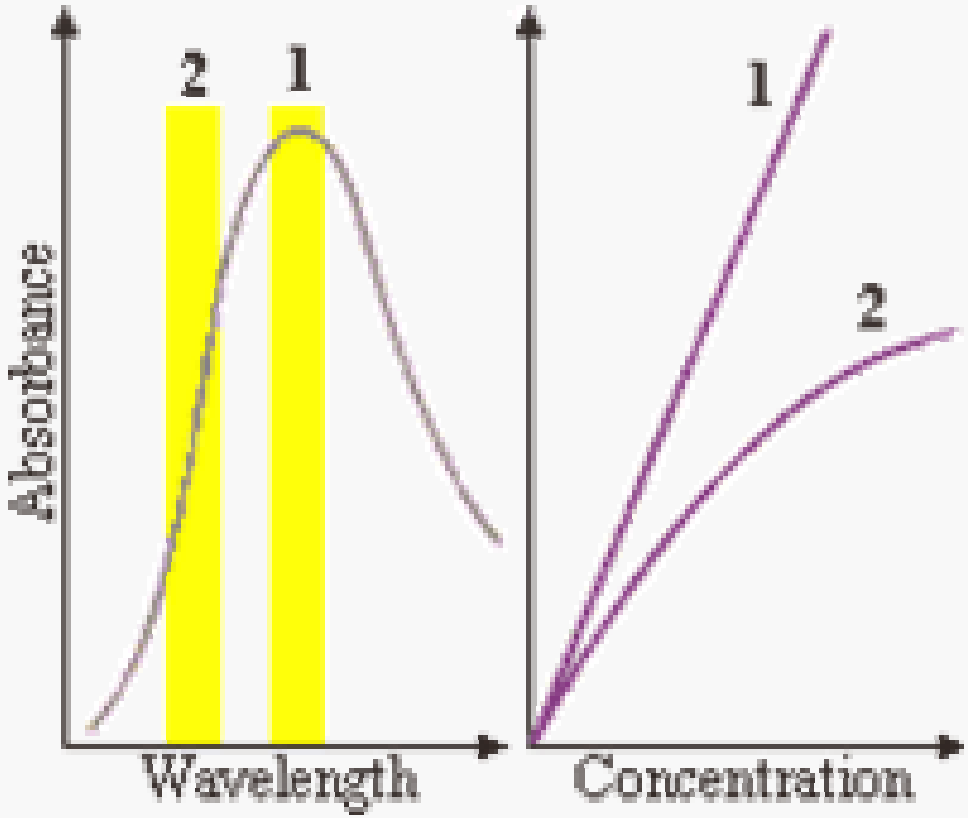
1) Only interaction between radiation (light) and the absorber (sample) is absorption. This breaks down if reflection and scattering are not compensated for. Also breaks down if the absorbed radiation is reemitted as fluorescence (not normally a problem) or if stray light in the instrument reaches detector

Assumptions made in deriving Beer's Law:

2) Monochromatic radiation – in reality this condition is only approximated, instrument measures a narrow band of radiation

ε varies with λ so the best place to measure A is at 1 where A is nearly constant with λ

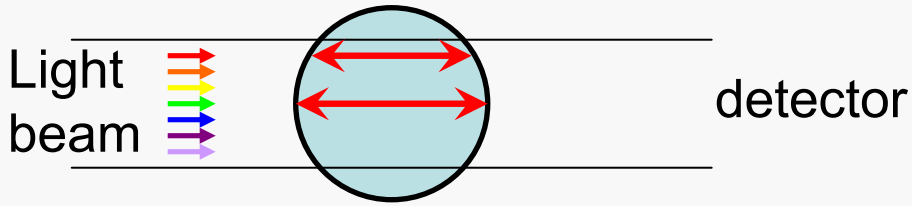
Measurements at 2 suffer from the variation in ε over the bandwidth



Assumptions made in deriving Beer's Law:

3) Pathlength is the same over the volume being measured – this becomes a problem with round cells

λ varies across the bandwidth so some wavelengths pass through more solution than others (b varies)

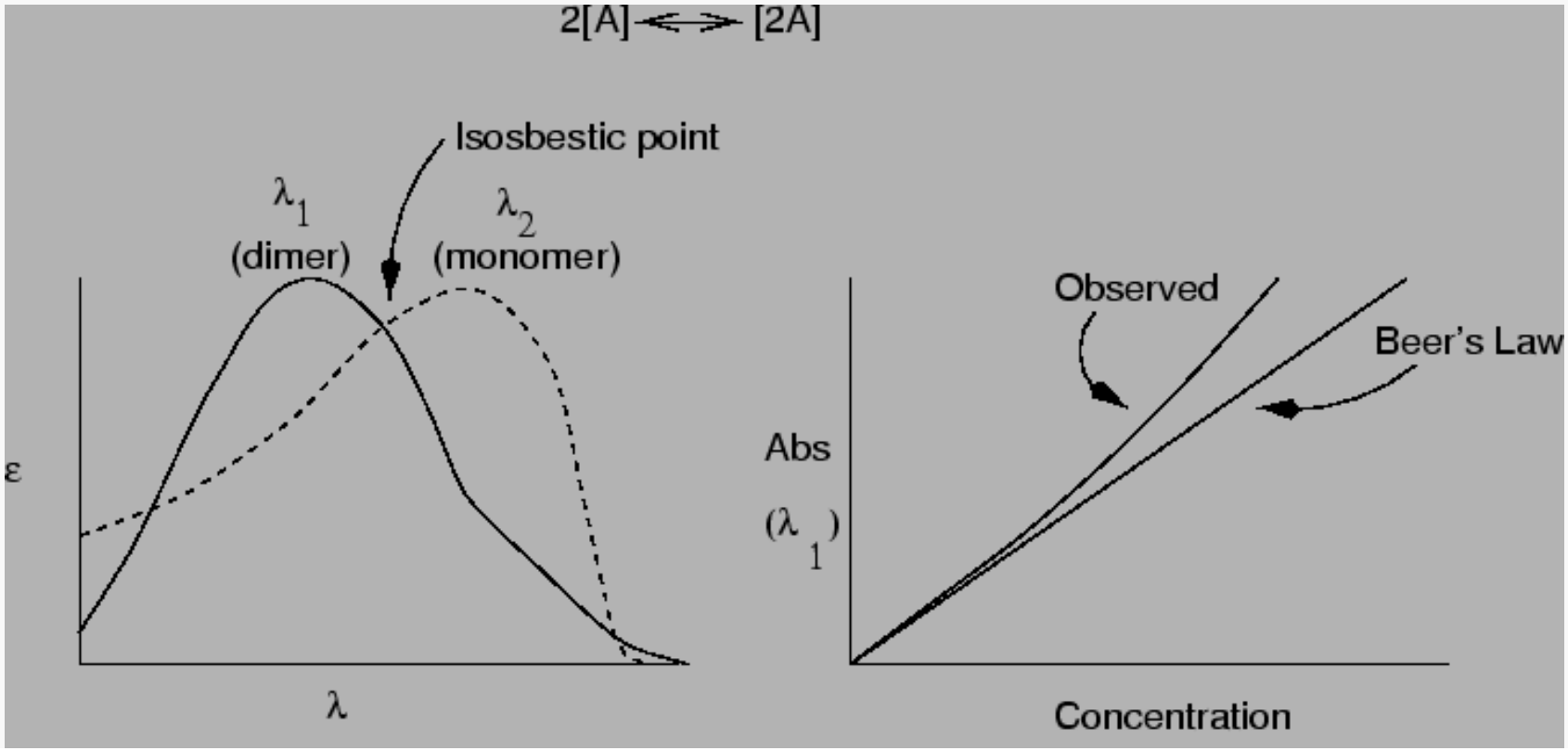


Round cell

The consequence is the same as for polychromatic radiation = curved response

Different components of the incident beam are absorbent with different efficiencies

4) The nature of the absorber does not change with concentration – a variety of effects can cause this assumption to break down, e.g. dimerization, acid-base or complexation equilibria



All of the above mentioned deviations from Beer's Law (or instrumental deviations) are really only deviations in the sense that the experimental conditions deviate from the conditions that have been assumed in deriving Beer's Law

Beer's Law always holds

Types of Spectroscopy

- Absorption
  - Atomic – AA - not covered
  - Molecular
    1) UV-vis – electronic
    2) IR – vibrational
    3) Microwave – rotational
    4) NMR (radiowave, MHz) – nuclear spin
    5) ESR/EPR (GHz) – electron spin

Types of Spectroscopy

Emission

- Atomic – AE & AF - not covered
- Molecular
  1) Fluorescence
  2) Phosphorescence
  3) Chemiluminescence

} Luminescence (UV-vis region)

Types of Spectroscopy ?

- Scattering
  - Raman spectroscopy – infrared region
  - Turbidimetry – UV-vis region
  - Nephelometry – UV-vis region
- Index of Refraction
  - Refractometry
  - Optical Rotatory Dispersion

Ultraviolet – Visible – Infrared Instrumentation

- Focus our attention on measurements in the UV-vis region of the EM spectrum
- Good instrumentation available
- Very widely used techniques
- Longstanding and proven methods
- IR instrumentation will be considered from time to time particularly when there are similarities to UV-vis

Absorption measurements require:

1) source of radiation

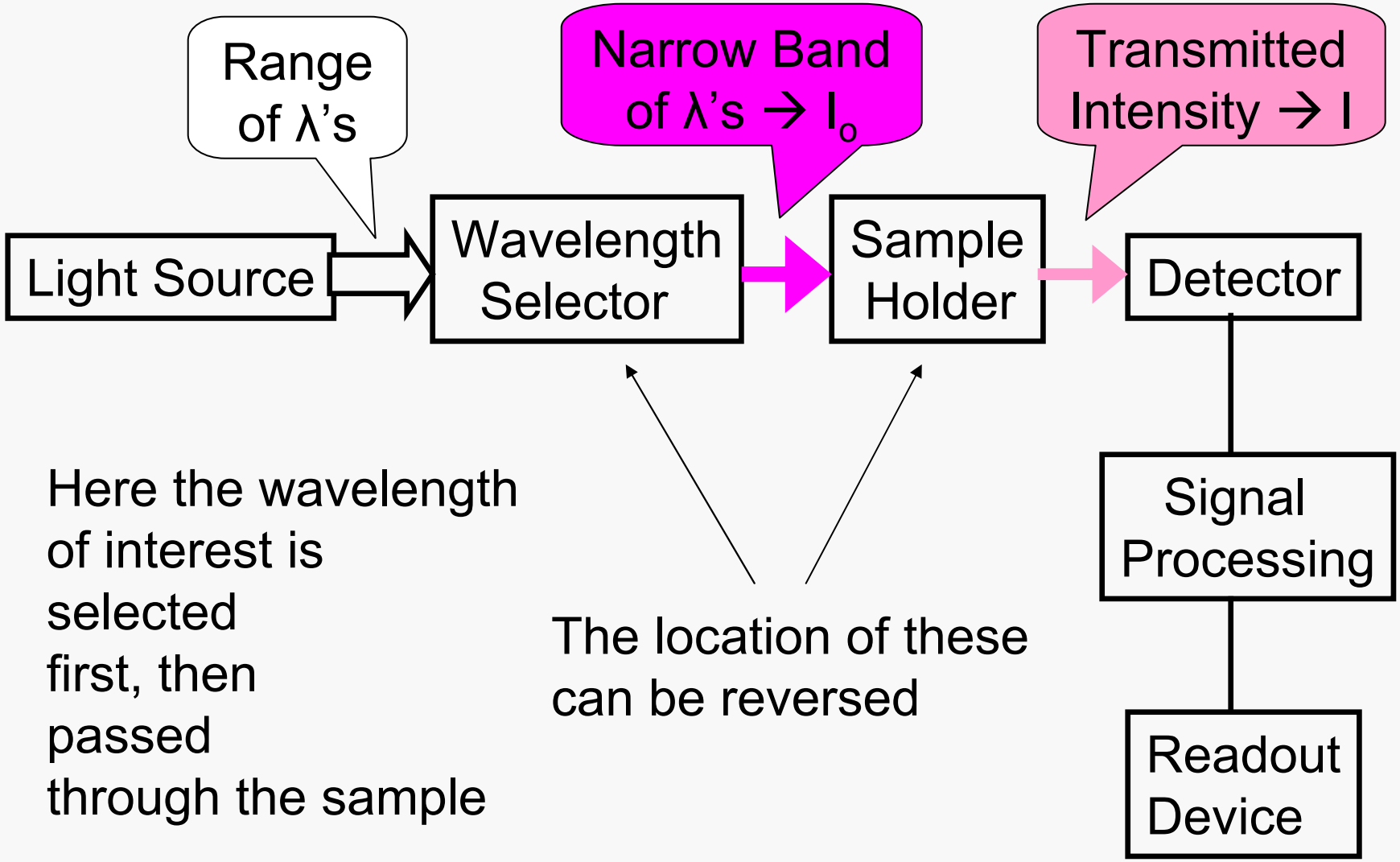
2) device for dispersing radiation into component wavelengths

3) a means of putting sample into the optical path, i.e., cell

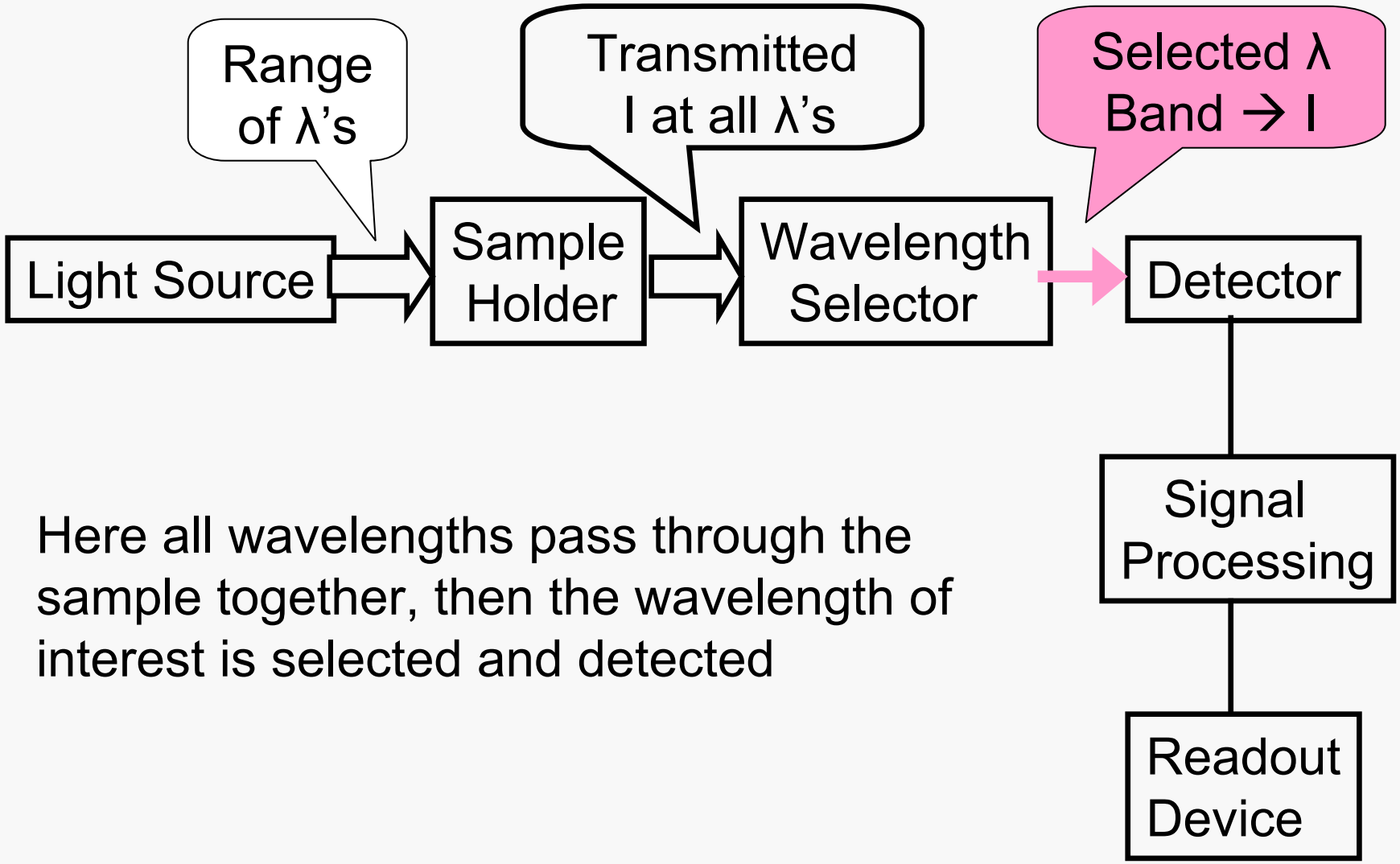
4) Detector to convert the EM to an electrical signal

5) readout device or circuitry, i.e., meter, computer, recorder, integrator, etc.

Block diagram of instrument for absorption



Block diagram of instrument for absorption



Here all wavelengths pass through the sample together, then the wavelength of interest is selected and detected

Emission measurements require:

1) means of exciting emission i.e., way of populating upper energy level which spontaneously emits

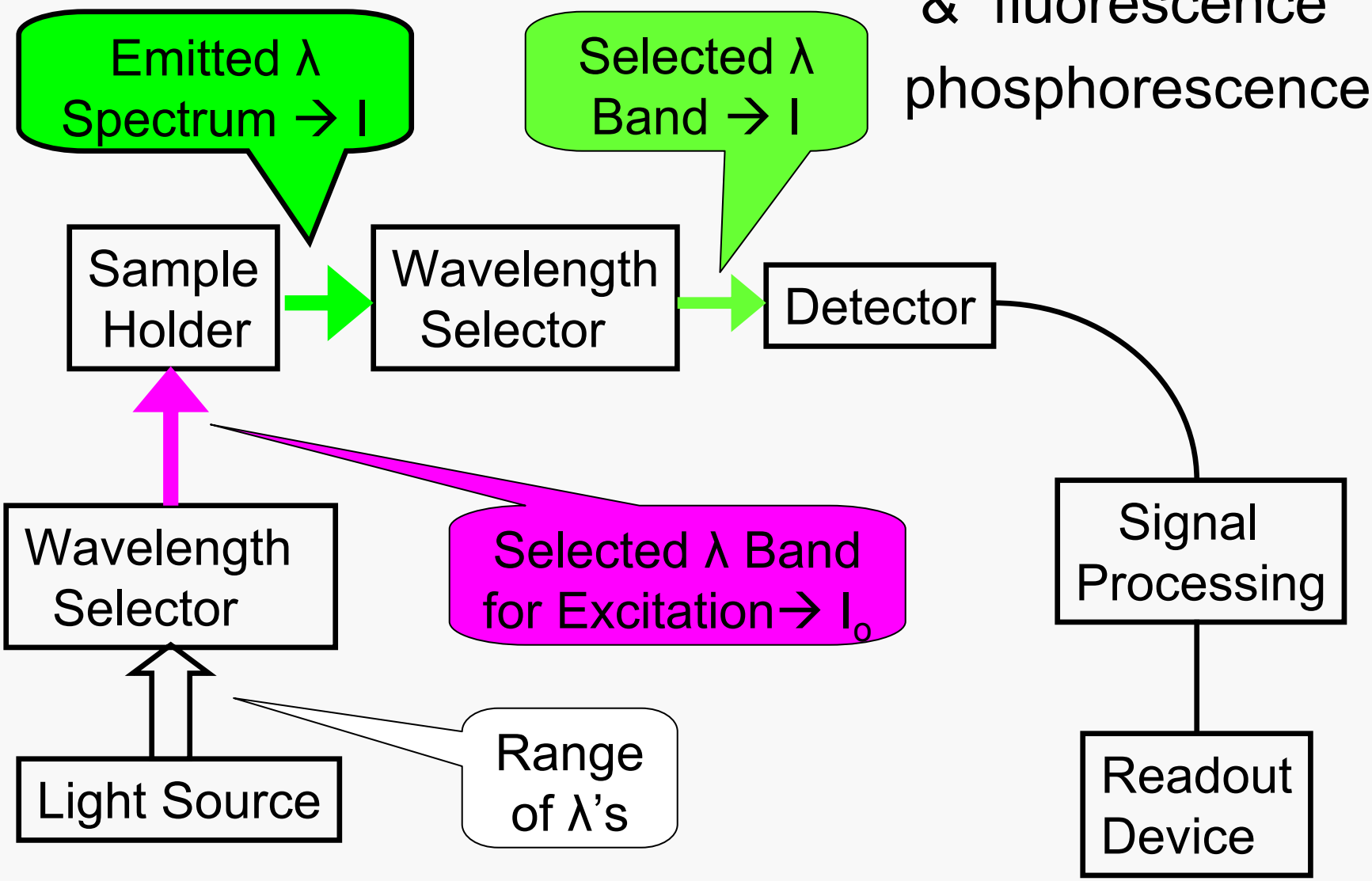
2) device for dispersing radiation into component wavelengths

3) a means of putting sample into the optical path, i.e., cell

4) Detector to convert the EM to an electrical signal

5) readout device or circuitry, i.e., meter, computer, recorder, integrator, etc.

Block diagram of instrument for emission i.e., & fluorescence phosphorescence



- The requirements for the various components used in different instruments change with the type of spectroscopy as well as for different kinds of measurements within a type of spectroscopy
- We will consider the components separately then combine them to make the overall instrument
- And finally look at the measurements with regard to theory and practice

Sources – important characteristics

1) Spectral distribution i.e., intensity vs. λ
   (continuum vs. line sources)
2) Intensity
3) Stability – short term fluctuations
   (noise), long term drift
4) Cost
5) Lifetime
6) Geometry – match to dispersion device

I) CONTINUUM SOURCES

1) **<u>Thermal radiation</u>** (incandescence) – heated solid emits radiation close to the theoretical "Black Body" radiation i.e., perfect emitter, perfect absorber

Behavior of Black Body

- Total power ~ T^4 therefore need constant temperature for stability when using incandescent sources
- Spectral distribution follows Planck's radiation law